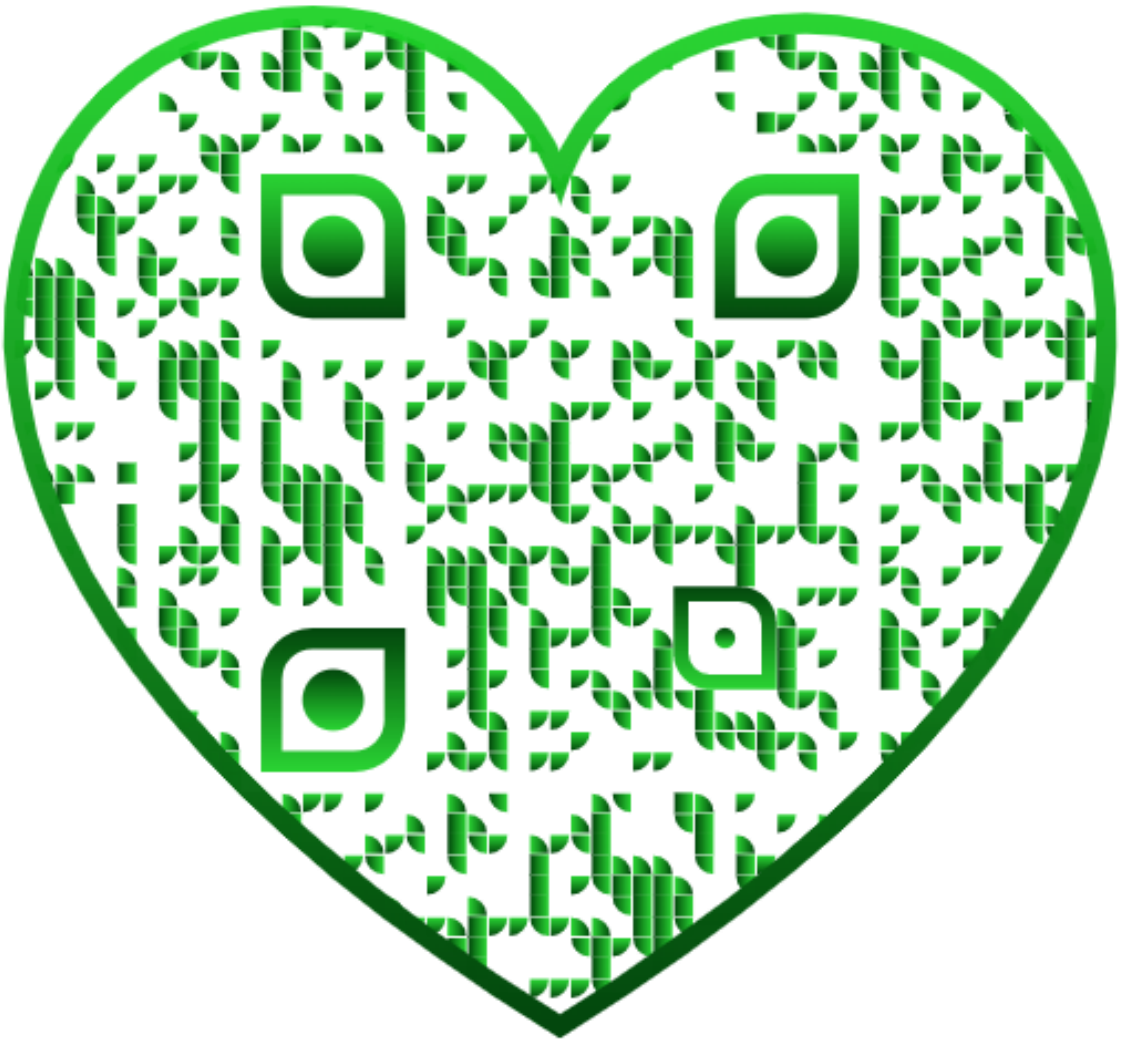
6. Sterile Products



Introduction

- The manufacture of sterile products is the most difficult of all pharmaceutical production activities to execute.
- The production of sterile products requires:
  - Fastidious design
  - Operation
  - Maintenance of facilities and equipment

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- Poorly controlled processes can expose the patient to an unacceptable level of contamination
- Sterile products are manufactured **aseptically**.

Aseptic Processing?

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- **Sterility:**
  - In absolute terms, sterility refers to a situation in which contamination is completely absent in a given product!!!
- **Asepsis:**
  - "A state of control attained by using an aseptic work area and performing activities in a manner that precludes microbiological contamination of the exposed sterile product"

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- Asepsis is the practice to reduce or eliminate contaminants from entering the manufacturing field.
  - Ideally, a "sterile" field, that is free of contaminants, is a situation that is difficult to attain.
  - However, the goal is elimination of infection, not sterility.

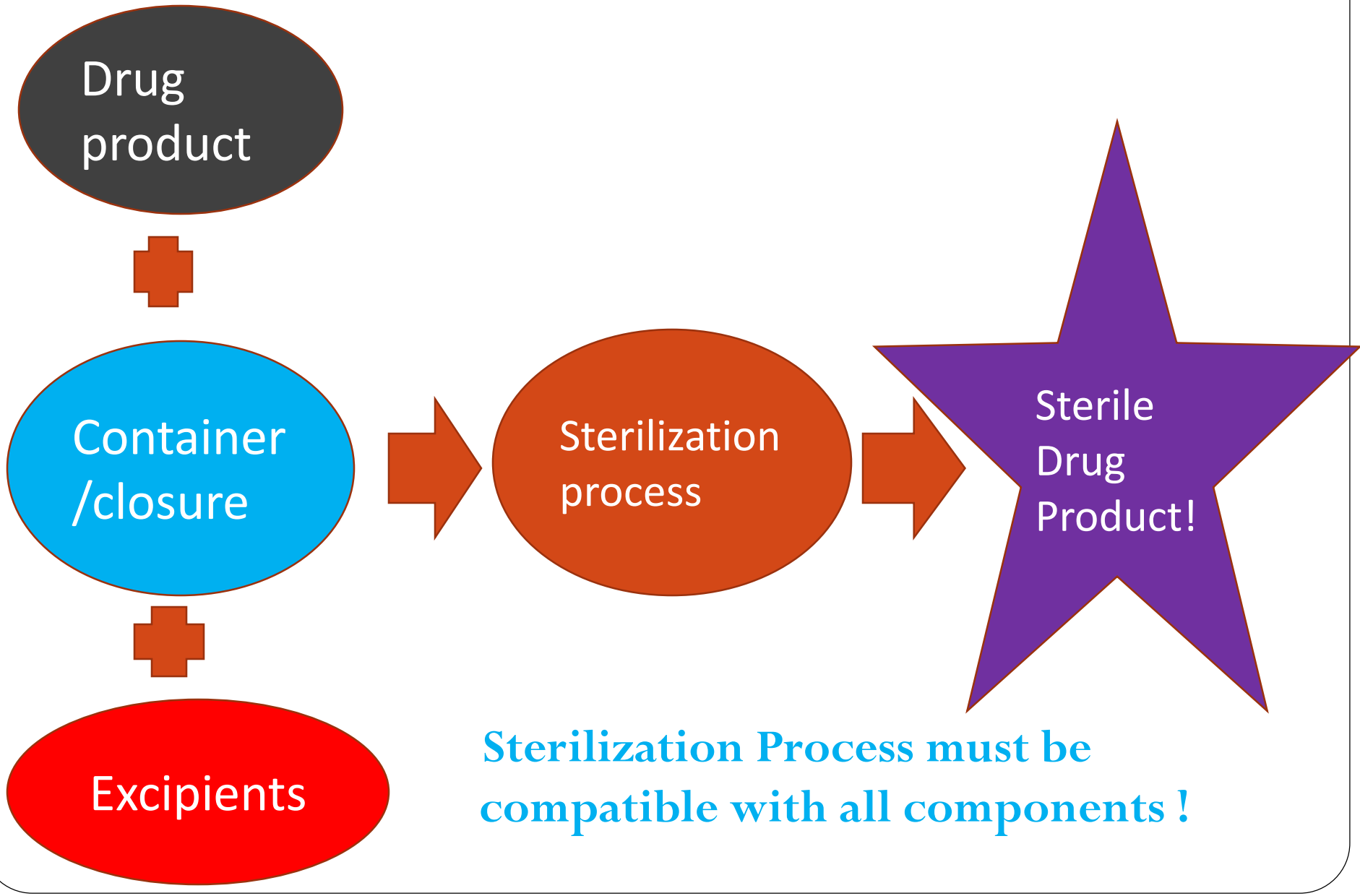
"Sterile drug manufacturers should have a keen awareness of the public health implications of distributing a non-sterile product. Poor CGMP conditions at a manufacturing facility can ultimately pose a life-threatening health risk to a patient."

FDA Guidance "Sterile Drug Products Produced by Aseptic Processing Current Good Manufacturing Practice" 2004.

Producing sterile drug products

- Sterile products can be produced either by Terminal sterilization or Aseptic processing.
  - **Terminal sterilization:**
    - Product containers are filled and sealed under high-quality environmental conditions designed to minimize contamination, but not to guarantee sterility.
    - Product in its final container is subject to a sterilization process such as heat or irradiation.

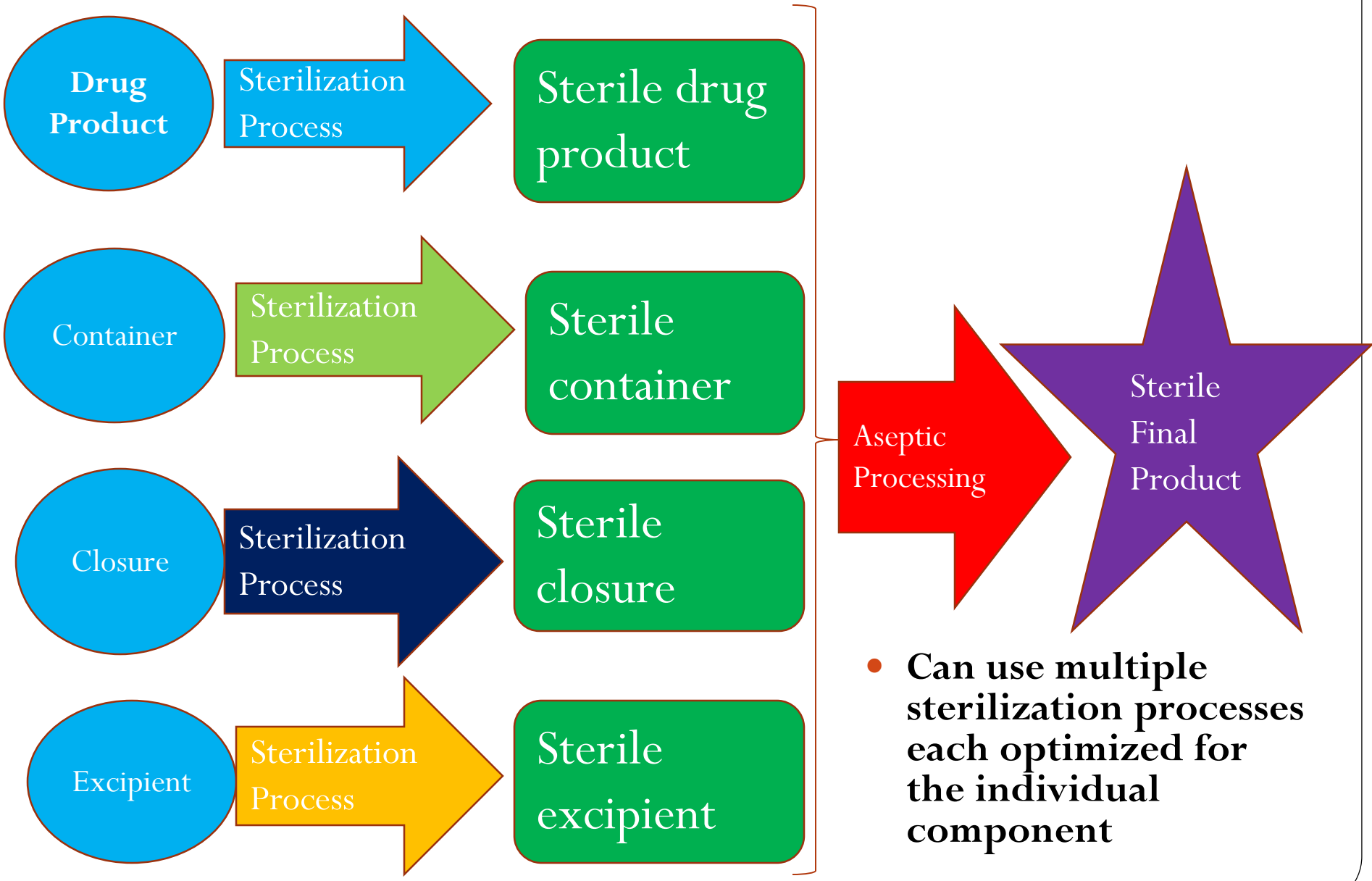
Terminal Sterilization



Sterilization Process must be compatible with all components !

- **Aseptic processing:**
  - Drug product, container, and closure are subject to sterilization separately, and then brought together.
  - Because there is no process to sterilize the product in its final container, it is critical that containers be filled and sealed in an extremely high–quality environment.

Aseptic Processing



- **Can use multiple sterilization processes each optimized for the individual component**

- The four pillars of a robust aseptic process :
  - Personnel training & monitoring
  - Environmental monitoring
  - Facilities design & HVAC validation
  - Process simulation (media fills)

- Avoiding contamination means knowing the potential sources of contamination:
  - Personnel
  - Equipment
  - Air/liquids
  - Drug product
  - Containers/closures
  - Outside environment

Viable & Nonviable Particles

- Particles of dust, fibers, or other material are suspended in the air and may contaminate product.
- These particles may, or may not, contain living organisms (bacteria and their spores).
- The more particles in the air surrounding the product the more likely the product will be contaminated with those particles.

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- Sneezing produces 100's of thousands of aerosol droplets that can then attach to dust particles.

- In the absence of any filtration system these particles which may contain bacteria spores, or viruses may be present in the air for weeks.

- Humans and bacteria:
  - Over 200 different species of bacteria are found associated with humans.
  - Bacteria are found in the intestines, eyes, nose, mouth, hair and skin.
  - Dry skin can have 1000's of microbes/mm^2

- **Would you let these people into your processing area?**

'Sick people aren't the only source of contamination!'

- Endotoxins:
  - Pyrogenic (fever inducing) substances (e.g. lipopolysaccharides) present in the bacterial cell wall.
  - Endotoxin reactions range from fever to death.
  - Extremely heat stable - recommended conditions for inactivation are 180 ^{O}C for 3 hours.

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- Spores:
  - Why are they a major challenge?
  - Heat alone will not inactivate….
- Viral Contamination:
  - Viruses are small (nm) non-living entities that “hijack” the machinery of a host cell.

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"The skin is home to a virtual zoo of bacteria" Martin J. Blaser

- If people are a major source of contamination, how do we avoid contaminating the product while we processing it?
- 1st step – eliminate the source of contamination!
  - "A Well designed, maintained, and operated aseptic process minimizes personnel intervention. As operator activities increase in an aseptic processing operation, the risk to finished product sterility also increases."

- 2nd Step - Reduce the risk of contamination through:
  - Sterile barriers
  - Aseptic technique
  - Environmental monitoring

- Gowning

Cap
(under hood)
Safety
glasses
Hood
Face mask
Gloves
Coveralls
Boots
Shoe
covers
(under boots)

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- Gowning Qualification:
  - Written (photographic) procedures describing methods used to don each gown component in an aseptic manner and the creation of barriers by overlapping gown components.
  - Initial training and periodic assessment.
- Environment monitoring:
  - Surface monitoring
  - Touch or contact plates

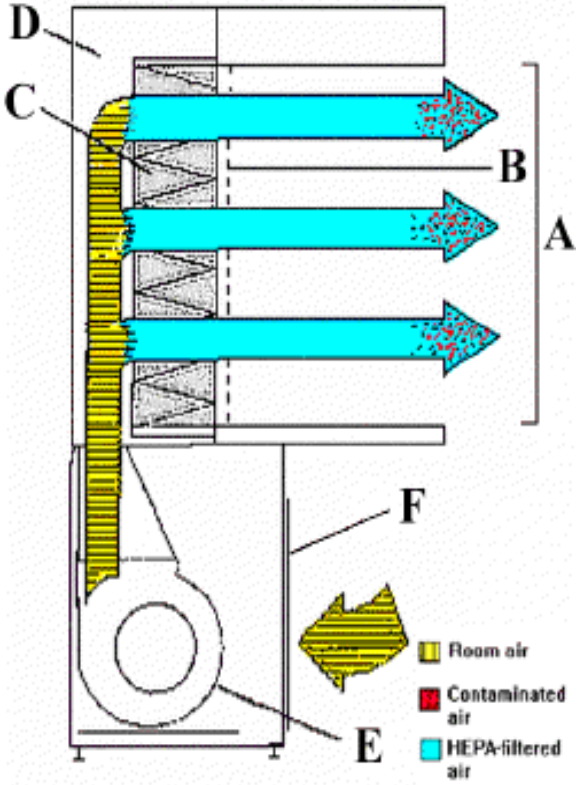
Aseptic Technique

- "Contact sterile materials only with sterile instruments"
- Sterile instruments should be held under appropriate conditions between uses and placed in sterile containers
- Operators should not contact sterile products, containers, closures, or critical surfaces with any part of their gown or gloves.

- During aseptic processing:
  - Keep the entire body out of the path of unidirectional airflow
  - Unidirectional airflow design is used to protect sterile equipment surfaces, container-closures, and product.
  - Disruption of the path of unidirectional flow in the critical area can pose a risk to product sterility.

Figure 9A. The horizontal laminar flow "clean bench"

A. front opening B. supply grille
C. supply HEPA filter
D. supply plenum
E. blower
F. grille



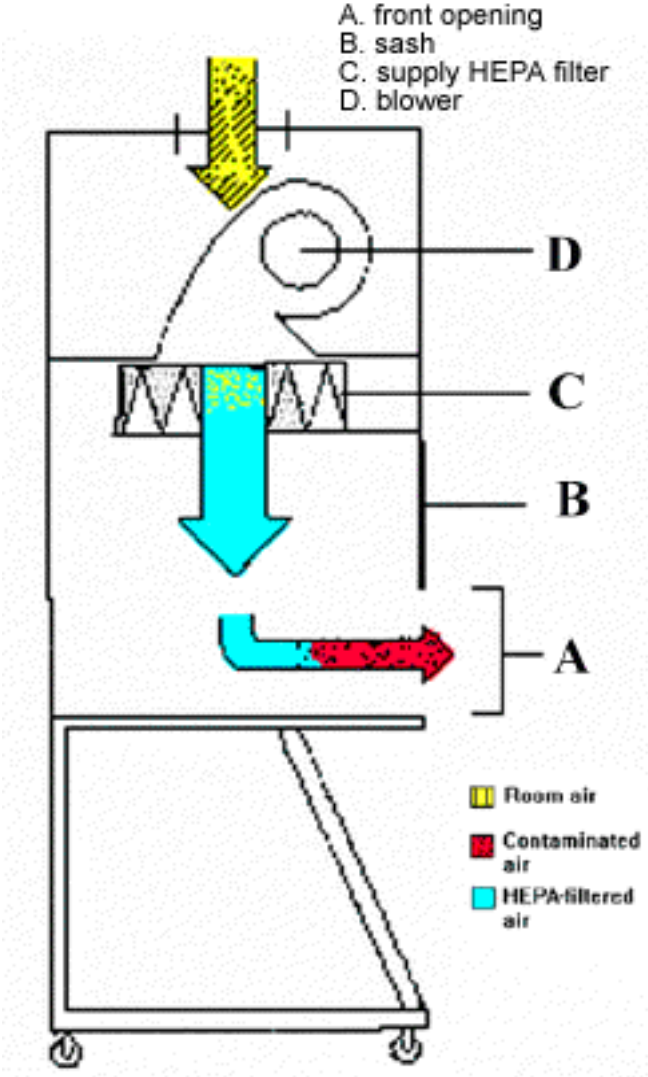
Horizontal airflow

Unidirectional airflow

The operator should never come between the air source and the product.

Figure 9B. The vertical laminar flow "clean bench"

A. front opening
B. sash
C. supply HEPA filter
D. blower



Vertical airflow

- Approach a necessary manipulation in a manner that does not compromise sterility of the product

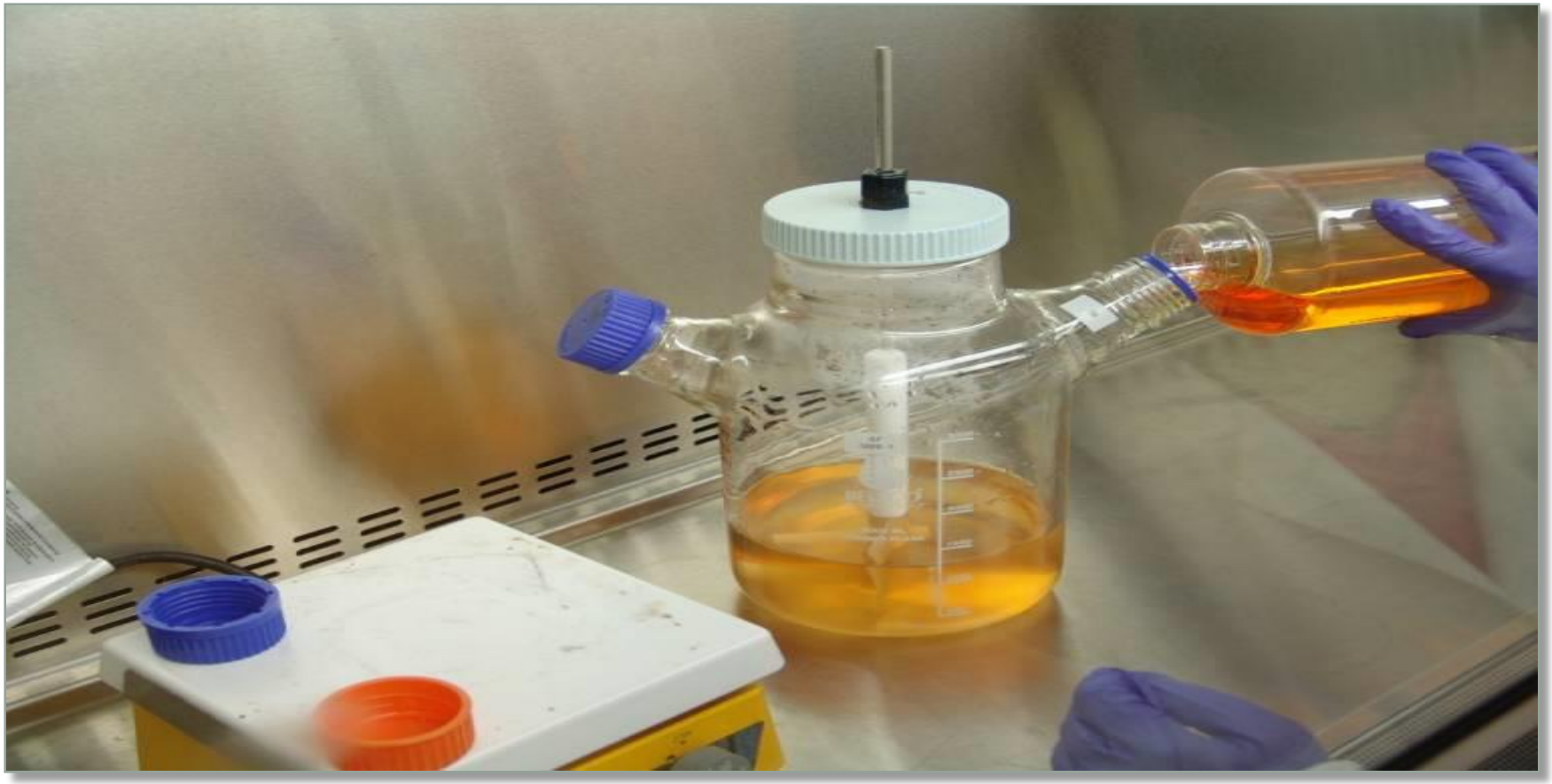
  - Proper aseptic manipulations should be approached from the side and not above the product (in vertical unidirectional flow operations).

  - Operators should refrain from speaking when in direct proximity to the critical area.

What's wrong with this picture?

Correct

What's wrong with this picture?



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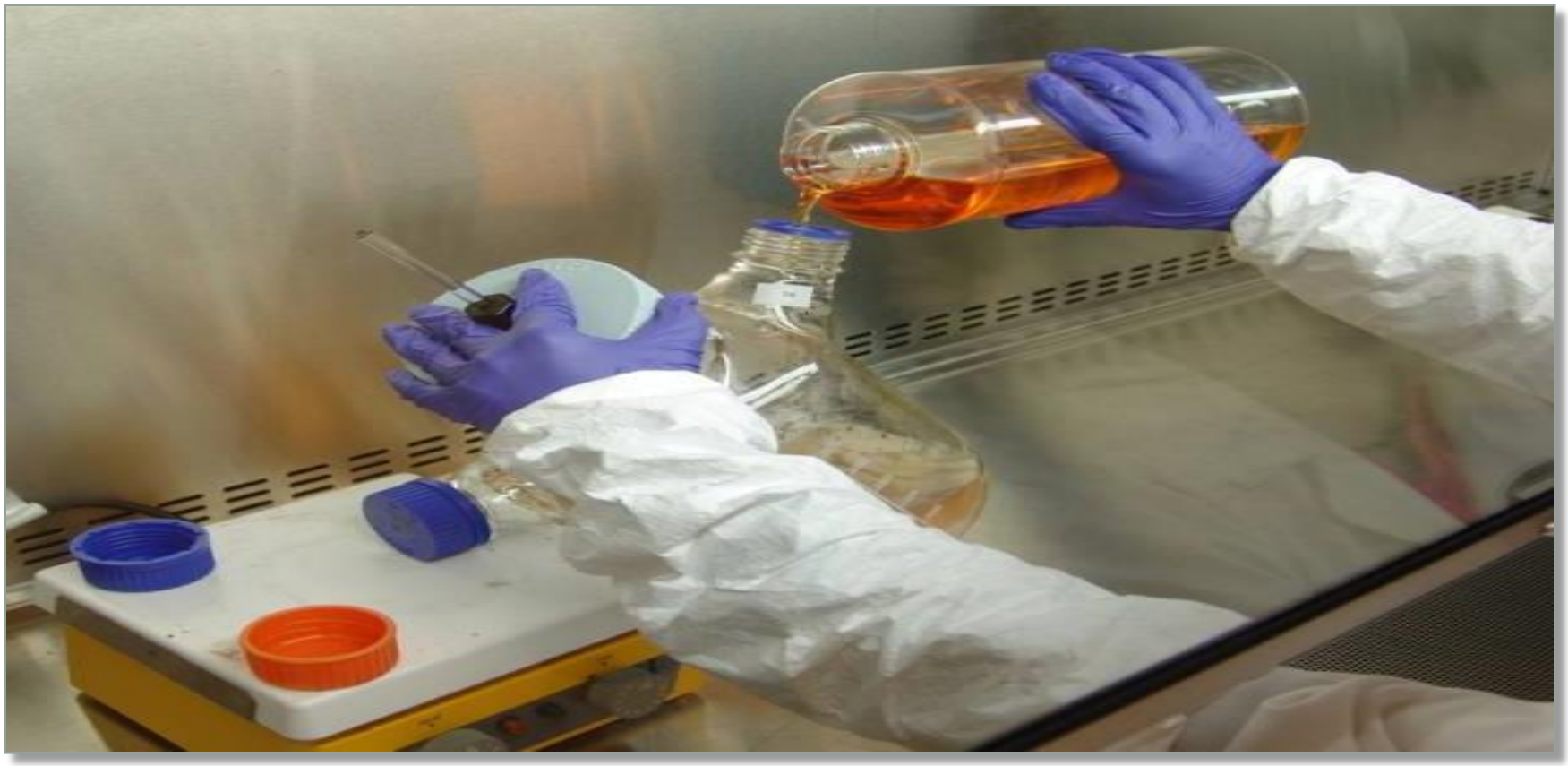

- Personnel: Hygiene
  - Avoid clean rooms when ill
  - Frequent bathing and shampooing
  - Avoid getting sunburned
  - Avoid cosmetics such as face powder, hair sprays, perfumes and aftershave
  - Clothing should be clean, non-frayed
  - Avoid smoking

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- Use of disinfectants is a pre-requisite in most aseptic processing lines.
- Isopropyl Alcohol (70%)
  - Powerful disinfectant and antiseptic.
  - Mode of action: denatures proteins, dissolves lipids and can lead to cell membrane disintegration
  - Effectively kills bacteria and fungi
  - But does not inactivate spores.

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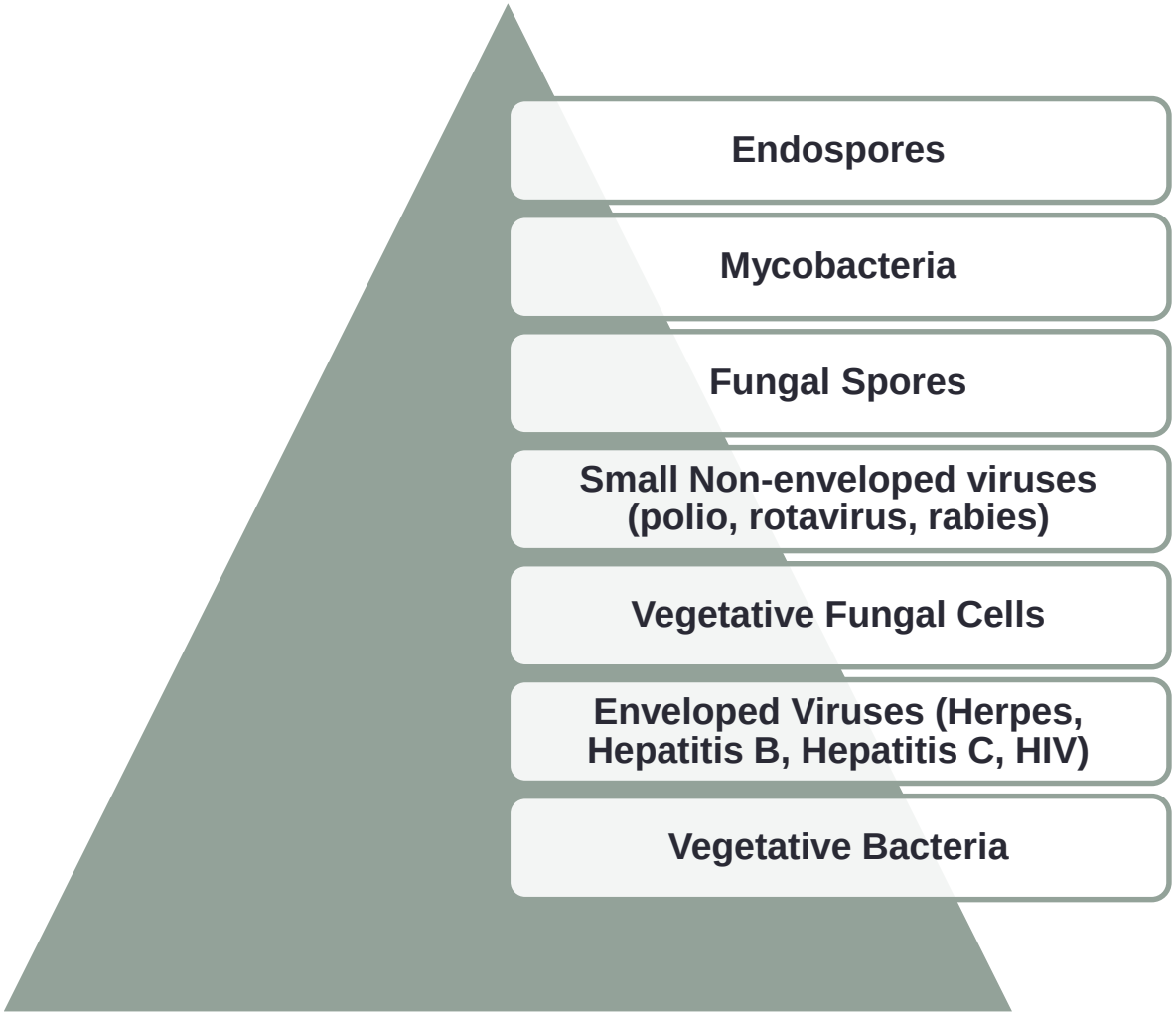
- The disinfection program should include the use of a sporicidal agent used according to a written schedule and when environmental data suggests presence of spore forming agents (*Baccilus spp.*).

Most Resistant



Least Resistant

Other disinfectants:

- Glutaraldehyde
- Formaldehyde
- Other aldehydes
- Chlorine-releasing agents
- Iodine and iodophors
- Peroxygens
- Ethylene oxide
- P-Propiolactone

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Equipment and bacteria

- Even seemingly smooth surfaces can harbor bacteria.
- Critical surfaces
  - Surfaces that may come in contact with or directly affect a sterilized product or its containers or closures.
  - Critical surfaces are rendered sterile prior to the start of the manufacturing operation, and sterility is maintained throughout processing.

- Isolators

- Advantage:
  - No direct contact between operator & product
  - Critical that meticulous aseptic practices be followed including the use of sterile tools for manipulations

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- Open isolator
  - Require laminar airflow over critical areas
  - Use pressure differential to insure separation of critical area from external environment.
  - Local protection of opening to guard against turbulent airflow and pressure waves that could compromise critical area

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- Isolator Decontamination
  - Vaporized agents often used, E.g. Glutaraldehyde & Formaldehyde
  - Use indicator organisms to demonstrate effectiveness of decontamination
    - Biological indicator should be placed in multiple, justified locations throughout the isolator
    - Hard to reach areas (between fingers on gloves) should be addressed
    - Must show that defined concentration of decontamination agent is uniformly reached in validation studies

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Environmental Monitoring

"In aseptic processing, one of the most important laboratory controls is the environmental monitoring program"

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- The key for all aseptic processes is a “high quality environment”

- The goal of the environmental monitoring program is to provide meaningful information on the quality of the aseptic processing environment during production as well as environmental trends and historical data.

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- What should an environmental monitoring program cover and how?
  - All production shifts
  - Air, floors, walls, equipment surfaces including critical surfaces.
  - The location of surfaces to be sampled, timing, and frequency of sampling should be specified in writing.
    - Sufficient detail to allow reproducible sampling

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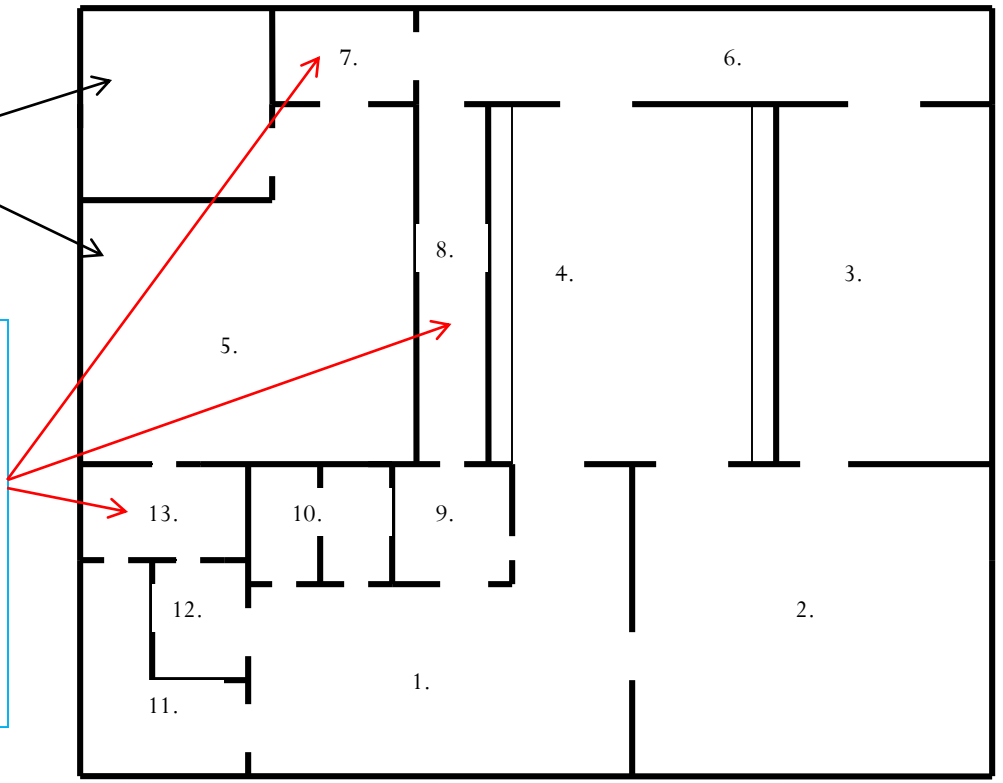
Where do we sample?

Critical (processing) areas

Sampling of adjacent classified areas (aseptic corridors, gowning rooms, etc) will provide trend
data and may help identify sources of contamination.

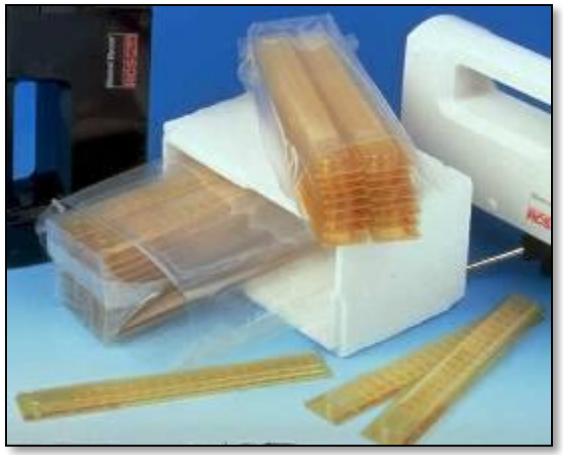


- Environmental monitoring SOP's include:
  - Frequency of sampling
  - When the samples are taken (during or at conclusion of aseptic operations)
  - Duration of sampling
  - Sample size (surface area, air volume)
  - Specific sampling equipment and techniques
  - Alert and action levels
  - Appropriate response to deviations from alert or action levels

- Particulate Air Monitoring

Use of remote systems recommended in laminar flow areas

- Viable Microbial Air Monitoring
  - Active Air Monitoring, RCS Plus
  - Passive Air Monitoring, Settling Plates



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- Media must be qualified to support growth of USP indicator organisms
  - To support growth of bacteria and fungi
- Upper and lower limits for incubation 20-35^{O}C, and not less than 14 days (if two temps 7 days at each temp)

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- Microbial identification should extend to the species level.
- Routine identification can be done using traditional phenotypic and biochemical techniques.
- Genotypic techniques are suggested for failure investigations.

Facilities

- Establishing and maintaining an aseptic environment
  - Use clean-rooms of various classes to establish an aseptic area.
  - Clean rooms use combinations of filtration, air exchange, and positive pressure to maintain "clean" environment.
  - Lower quality clean areas should not be placed next to high quality areas.

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- Facility Design: Clean Area Classification
  - For the manufacture of sterile medicinal products there are normally 4 grades of clean areas.

	at rest		in operation	
Grade	maximum permitted number of particles/m3 equal to or above			
	0.5 µm	5 µm	0.5 µm	5 µm
A	3 500	0	3 500	0
B	3 500	0	3 50000	2 000
C	350 000	2 000	3 500000	2 0000
D	350 0000	2 000	Not defined	Not defined

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- **Grade A**
  - The local zone for high risk operations, e.g. filling zone, stopper bowls, open ampoules and vials, making aseptic connections.
  - Normally such conditions are provided by a laminar air flow work station.
  - Laminar air flow systems should provide a homogeneous air speed of 0.45 m/s ± 20 % (guidance value) at the working position.

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- **Grade B**
  - For aseptic preparation and filling, this is the background environment for grade A zone.
- **Grade C**
  - Clean area for carrying out less critical stages in manufacture of aseptically prepared products.
- **Grade D**
  - Clean area for carrying out less critical stages in manufacture of aseptically prepared products.

Grade C clean room

Grade A clean room

- General Clean-room design:
  - HEPA/ULPA filters on ceiling
  - Exhaust vents on floor
  - Airlocks and interlocking doors to control air balance
  - Seamless and rounded floor to wall junctions
  - Readily accessible corners
  - Floors, walls, and ceilings constructed of smooth hard surfaces that can be easily cleaned
  - Limited equipment, fixtures and personnel
  - Layout of equipment to optimize comfort and movement of operators

- **HEPA Filters**
  - High Efficiency Particulate Air
  - Minimum particle collection efficiency:
    - 99.97% for 0.3μm diameter particles.
  - Disposable
  - Filter made of **pleated borosilicate glass**

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- **Pressure Differentials**
  - Used to maintain airflow in the direction of higher cleanliness to adjacent less clean areas
  - A minimum of 10-15 Pascals should be maintained between the aseptic area and an adjacent rooms with differing clean room classifications (doors open)

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- **Air Lock**
  - Permits the passage of objects and people into a clean room.
  - Consists of two airtight doors in series which do not open simultaneously.
  - Spray down materials with 70% IPA before placing in the airlock

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- **Material NOT permitted in a Clean room**
  - Fiber-shedding materials such as cardboard and paper
  - Cardboard packaging must be removed and items placed into non-cardboard containers.
  - Wood (i.e. wooden pallets)
  - Undesignated charts

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- **Cleaning**
  - 1. Vacuum all accessible surfaces
  - 2. Wipe surfaces with a cleaning solution
  - 3. Mop floors using a lint free polyester mops attached to stainless steel handles
  - Water should be changed FREQUENTLY